

salmon-nets (in the latter especially in easterly gales, which rendered the water muddy) ; in one case, indeed, the net could not be pulled up off the east rocks, from the great weight of the captured lumpsuckers (estimated at several tons), and it was ruptured. They are only used along with fish-offal for manure.

Pennant's observation with regard to the tenacity with which an adult clings by its sucker to a pail full of water has been found to be quite accurate. The whole can be lifted by seizing the fish, and a greater weight than 43 pounds (which was that of pail and water) could readily be raised in this manner.

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X.—*On the Presence of Oleaginous Spheres in the Yolk of Teleostean Ova.* By EDWARD E. PRINCE, St. Andrews Marine Laboratory.

OF the 9000 or 10,000 species of osseous fishes known to zoologists the eggs of not more than 80 have been obtained and determined. This comparatively small number indeed includes several species whose ova have been discovered only within the last twelve months by Prof. McIntosh at the St. Andrews Marine Laboratory, and are therefore new to science. Quite a large proportion of these eggs are characterized by the presence in the yolk of large refringent masses, the so-called oil-globules. These structures have long been familiar to embryologists, and they constitute a prominent feature in those Salmonoids whose development is more completely known than that of any other group of Teleosteans. Yet the significance and function of these bodies seems to be little understood, or, more truly, seems to be wholly misunderstood. Of course all fish-ova have oily elements in their protoplasm, some cholesterin being constantly present, with other fatty matters, in addition to myosin and the usual derivatives of albumin; but these elements, when they can be detected optically, are microscopic, and, being distributed as minute vesicles all over the vitellus, strikingly differ from the large globules here considered. Not only in size, but in colour, situation, and relation to the rest of the ovum, and almost certainly also in chemical composition, these large spheres are distinguished from the microscopic fatty particles present in all ova. So well-marked and characteristic are

these spheres that the species to which an ovum belongs may often be determined at a glance by their aid.

Struck by their diagnostic significance, Agassiz and Whitman divided pelagic eggs into two great divisions, those which are provided with one or more oil-globules and those which are not so distinguished\*. Such a division has, however, little value, as the occurrence of these large globules is apparently most erratic—the ova of closely-allied species exhibiting the utmost diversity in this respect. Indeed the American observers themselves describe an ovum of a Pleuronectid (*Pseudorhombus oblongus*) showing a large oily sphere, a structure not present in the egg of any other species of flounder known to zoology. The ova of the Gadoids, too, are marked by the absence of such structures; yet a remarkable exception has been recently found at St. Andrews, viz. the hitherto undescribed pelagic egg of the ling (*Molva vulgaris*), which exhibits a single oleaginous sphere of a pale green tint. Additional exceptions are furnished by the closely-allied freshwater species, *Lota vulgaris* (the oil-globule in which, as described by Van Bambeke †, is almost precisely like that in the egg of the ling), as well as the eggs of the American *Brosmius* ‡ and *Motella mustela* §, while the ova of all other Gadoids at present known lack this marked feature, no large globule being present in the case of the cod, haddock, whiting, bib, &c. Again, we find amongst freshwater forms that the ova of the salmon, trout, and grayling have large rufous-tinted spheres enveloped in the deutoplasm or yolk, whereas in other forms which inhabit the same waters and deposit their ova in similar situations, such as the pike ||, tench, roach ¶, &c., no such globules are present. A classification of ova founded on the presence or absence of these spheres presents a strange medley—the two lists bringing together widely separated species and placing side by side fishes with pelagic and demersal ova, and most diverse freshwater and marine forms. An undoubted specific value belongs to these spheres; but no generic or wider diagnostic significance can be attributed to them. Certainly the interpretations which the presence of these globules has hitherto received are not only unsatisfactory, they are undoubtedly

\* 'Studies from Newport Marine Laboratory.—XVI. Dev. Oss. Fishes,' p. 2 (1885).

† "Recherches sur l'embryologie des poissons osseux," Mém. Cour. de l'Acad. Roy. de Belgique, tome xl. p. 5.

‡ U. S. Fish. Comm. Rep. 1882, p. 467.

§ G. Brook, Journ. Linn. Soc., Zool. vol. xviii. p. 298.

|| E. B. Truman, Month. Microsc. Journ. vol. ii. 1869, p. 188.

¶ Van Bambeke, *loc. cit.* pp. 2 and 13.

erroneous. Ryder attributes the buoyancy of certain pelagic ova to these structures, and, strangely enough, later observers have put forward the same view, notwithstanding the fact that the most familiar of Teleostean eggs, viz. those of the Salmonoids, possess such spheres in abundance, and yet are wholly destitute of the power of floating. In a list of twenty-two Teleostean ova without large globules, seventeen (or about 75 per cent.) are pelagic. On the other hand, about twenty-four species of Teleosteans are known to possess these globules, and fifteen of these are pelagic, a proportion not far removed from that furnished by the list just named. In other words, the pelagic eggs without large globules are about the same in number as those possessed of globules, so far as researches at present show; and to explain the buoyancy of floating eggs by the presence of these structures is a manifest fallacy\*. Moreover, large globules are present not only in demersal eggs which are littoral, *i. e.* deposited near shore, such as those of various species of *Cottus*, *Liparis*, *Gastrosteus*, &c., but ova brought up from some depth show their presence also, an example of great interest being the large non-floating egg of *Anarrhichas lupus*, which has been reared and studied at St. Andrews for the first time. From an examination of the ovaries of the catfish in February 1884 Prof. M'Intosh concluded that the ova were deposited on the bottom of the sea †, and they have proved to be so; yet they exhibit a single refringent globule of large size of precisely the same appearance as the globules in the familiar ovum of *Cyclopterus*, which is deposited between tide-marks. It is plain that while these globules are of less specific gravity than the remaining contents of the egg, as is shown by the fact that they always seek the upper side, whereas the germinal area descends to the lower side of the ovum, yet they do not produce buoyancy; nay, in demersal eggs these vesicles are even more abundant than in pelagic or floating eggs. Their function is plainly not hydrostatic. A second theory has been put forward by Van Bambeke, viz. that the globules have a nutritive function; and in speaking of the central globule in the egg of the burbot (*Lota vulgaris*) he says:—"Il n'est pas douteux que la gouttelette réfringente centrale remplace ici les éléments nutritifs qui, chez la tanche, vont s'accumuler sous le germe" ‡. He adds this important

\* *Vide* Prof. M'Intosh's observations, 'Nature,' vol. xxxi. p. 555; Ann. & Mag. Nat. Hist. 1885, vol. xv. p. 435 &c.

† Ann. & Mag. Nat. Hist. June 1885, p. 432, and 'Nature,' June 24, 1886.

‡ *Op. cit.* p. 6.

statement:—" Sur quelques œufs, j'ai vu une communication s'établir entre le germe et la gouttelette du globe vitellin, comme si le germe allait puiser à cette source de nutrition ;" and in the figure which he gives (pl. i. fig. 14) it is difficult to explain the existence of the column of protoplasm connecting the globule and the germ, except as indicating a trophic function, unless the ovum were abnormal, which it most probably was. Unfortunately the ovum in question was not fertilized, and the subsequent fate of the globule was not ascertained. In studying the complete development of the ling, gurnard, catfish, and other forms at St. Andrews unusually favourable opportunities were afforded for tracing the destination of the globular bodies, and the evidence gained strongly militates against Van Bambeke's theory that the germinal disc is nourished by them. Not only do they show no decrease in size and persist in the pendulous yolk for some time after the embryo is liberated, but the actual position of the globules in the early ovum is unfavourable to Van Bambeke's view.

As already stated, the normal position of the globules is constant, viz. in the upper segment of the ovum, at the vegetal pole, and they therefore occupy the region most distant from the germinal disc. In a small number of eggs, however, this is not the case, and a remarkable example described by Agassiz and Whitman\* is the ovum of *Cottus grænelandicus*, in which from ten to forty globules are more or less evenly scattered over the surface of the yolk. This ovum, strange to say, is pelagic; whereas all other Cottoids, so far as known, have demersal eggs, and all alike are abundantly supplied with large oil-globules. A similar condition occurs in the ovum of *Trachinus vipera*—in which the oil-globules according to G. Brook are "scattered over the upper hemisphere of the yolk, and lie between it and the vitelline membrane" or capsule †. The large globule in *Lota vulgaris* is central, but this position is very unusual, and it is perhaps permissible to suggest that Van Bambeke may have mistaken the apparent for the real position of this body. The globule always rises to the upper side of the egg, and when the latter is on the stage of the microscope, unless by very careful manipulation, the actual position of the vesicle cannot be made out. Viewed under the usual microscopic conditions, the oil-globule in *Motella*, *Trigla*, *Molva*, &c. appears to be central, when in reality it is not so. The oil-globule in truth occupies different

\* 'Studies from Newport Mar. Lab.—XVI. Develop. Oss. Fishes,' p. 7.

† G. Brook, Journ. Linn. Soc., Zoology, vol. xviii. p. 274.

situations in different species, occurring within the yolk mass or outside it, in the perivitelline space, or rather in a fossa or pocket indenting the surface of the yolk. Examples of the latter condition are afforded by the Gadoid ovum studied by Hückel, and by *Motella mustela*, *Lophius piscatorius*, *Molva vulgaris*, and other forms. Instead of being seated, however, in a depression or pocket lined by the cortical protoplasm of the vitellus, the large vesicles may lie within this protoplasmic layer, or rather in the albuminoid matrix of the yolk.

In *Gastrosteus*, *Liparis*, *Cottus*, *Cyclopterus*, and other demersal eggs the globules, which are very numerous, and collect together in a large group at the vegetal pole, are thus surrounded by yolk substance, which, however, has sufficient fluidity to permit free movement, and the mass of vesicles may be made to traverse all parts of the inner surface of the yolk cortex, by turning the egg about in various directions. An interesting American pelagic egg, *Temnodon saltator*, which exhibits a single globule only, is in like manner imbedded, and has apparently shifted to a position immediately beneath the germinal disc in the figures given by Agassiz and Whitman\*. Professor McIntosh has proved that the globule in *Trigla gurnardus* does not occupy a position in the perivitelline space, as some observers have stated, but freely moves through the deutoplasmic mass.

Though thus capable of transference from one region of the yolk to another, the normal position always is distal to the animal pole, and to this upper (vegetal) segment the globules invariably return when the rotated egg comes to rest. These vesicles in some ova seem to have less freedom of movement, and appear to be imprisoned by the surrounding matrix.

Thus E. Van Beneden speaks as follows of the ovum examined by him:—"The animal pole was always directed downwards, the vegetative pole upwards. I ascertained that in my eggs the position of the oil-drop was quite constant. It is always placed eccentrically, and invariably occupies a position in the vegetative hemisphere, but is immersed in the albuminoid substance which surrounds it on all sides. I have in vain endeavoured to explain to myself this fact by some peculiarity of structure in the protoplasm. I entirely failed to discover any trace of filaments connecting the oil-drop either with the surface of the vitellus, or with the germinal disc"†. It may be noted that the pseudopodial threads here referred to have been seen in *Gastrosteus spinachia*‡, *G. aculeatus*, *G. pun-*

\* 'Studies from Newport Mar. Lab.—XVI.,' plate iv, figs. 1 and 2.

† E. Van Beneden, Quart. Journ. Micr. Sci. vol. xviii. p. 44.

‡ Ann. & Mag. Nat. Hist. 1885, vol. xvi. p. 492.

*gitius*\*, *Tinca vulgaris* †, &c. In the salmon the globules are held in position by the coherent granular cortex of the vitellus; they are not, however, merely free vesicles defined by the surrounding matrix, but, as His notes, "sind je von einer Hülle protoplasmatischer Substanz umgeben" ‡. This enveloping coat is well seen in the gurnard, and it increases in thickness as development proceeds, being very uneven and imprisoning many small colourless vesicles, precisely as His figures in the case of the ovum of the salmon &c. §

Van Beneden has omitted to show any definite layer, and Lereboullet does not indicate it in the ovum of *Perca* ||, though it is improbable that in either case the globule is destitute of the limiting layer present in other forms. His figures, in the plates just referred to, connected and isolated globules in the eggs of the salmon, trout, and grayling, and discusses fully the character of the coherent granular protoplasm which clothes them ¶.

The precise chemical nature of these large globules is still involved in some uncertainty. That they are of an oleaginous nature cannot be doubted, though it is scarcely accurate with E. Van Beneden to describe a sphere of this kind as "a drop of oil or fat," for the investigations of Professors His and Miescher show its composition to be that of no known fat \*\*. If an ovum of the gurnard, for example, be treated with osmic acid, the minute vesicles scattered over the vitellus stain very rapidly and deeply, whereas the large globule is coloured slowly and more faintly—proving the former to be more emphatically oleaginous than the latter. The large globules exhibit a more or less brilliant translucency; they float in water and are soluble by ether, though, according to Miescher they reveal no more than a trace of phosphorus.

Their composition, while closely allied, is not identical with that of any of the fats, and they may best be associated with those remarkable derivatives of albumin, the lecithin-group. To that group His, indeed, refers them, though he confesses that strictly their nature is undecided. The association of these spheres with lecithin is a matter of extreme interest, for lecithin is a substance always present in cells of ova under

\* Phil. Trans. vol. clvii. (1867).

† Van Bambeke, *op. cit.* p. 2, and plate i. fig. 2.

‡ His, *Untersuch. über das Ei und die Entwickl. bei Knochenfischen*, 1873, p. 7.

§ *Op. cit.* Taf. i. figs. 1, 2, 4, 5, 11, and 12, and Taf. iv. fig. 38.

|| Lereboullet, *Mém. des. sav. étrang. t. xvii. p. 460*, and plate iii. figs. 3, 7, and 8.

¶ *Op. cit.* pp. 6, 7.

\*\* *Ibid.* p. 7.

developmental conditions. May not this fact throw light upon the significance of these globules? We have seen that their presence cannot be explained by resorting to a hydrostatic function, and there are great difficulties in the way of the nutritive theory. Is it not possible that they may have some ancestral significance? The distinctive coloration they exhibit is an interesting point, though it can give no aid in the matter. Nevertheless it is remarkable that the orange tint of these spheres, in the ovum of the Salmonoids, is precisely that which distinguishes the oily matter in the muscular tissue of the adult fish. The flesh of the common mackerel, the Spanish mackerel, and the gurnard, not to mention others, is regarded as somewhat oily, and the ovum of each of these fishes exhibits a large globule. The oleaginous matter in the flesh of the last-named fish is of the same tint as that of the sphere in the ovum. The globule in the ling is of a pale green hue; and in the allied form, the burbot, Van Bambeke describes it as "très-réfringente, d'une teinte jaunâtre"\*.

The globules in the fifteen-spined stickleback, and in certain Cottoids, are of an amber colour, but in many forms (e. g. *Cyclopterus*, *Cottus*, *Motella*, &c.) they are almost perfectly colourless. These features are of minor importance, however, compared with the fact that in the ova of so many species of Teleosteans large spheres of a substance closely connected with the lecithin-group should occur. Lecithin is peculiarly active in all embryonic development, and the possibility is suggested that, though the matter constituting these globules may be disproportionately large as compared with the volume of the vitelline mass, yet it was not always so. That the yolk-matter of the Teleostean ovum was once greater in bulk than it is now, is (in accordance with Balfour's view †) an accepted conclusion. If as the vitelline mass diminished the lecithin or similar fluid did not decrease in the same degree, globules would be formed precisely as we find them in so many Teleostean eggs. The amount is more than the necessities of development appear to require; and thus we find that during the growth of the blastoderm, and during the early stages of the embryo, these superfluous elements are not utilized and do not appreciably decrease in volume. They are enveloped by the blastoderm, and in the liberated embryo generally occupy a posterior position in the diminishing yolk, on the ventral surface of the young fish. Finally they disappear in the last stages of larval life by absorption; but up to that point retain the character of redundant and superfluous elements in the deutoplasmic mass.

\* *Loc. cit.* p. 5.

† *Journ. Anat. and Phys.* vol. x. p. 551.